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## A viscometric study of solubilized calfskin collagen using the ZIMM-CROTHERS viscometer

A recent investigation of the viscometric behavior of solubilized calfskin collagen was carried out in capillary flow viscometers having very long capillaries so as to give low rates of shear<sup>1</sup>. When values of shearing stress were plotted against rate of shear to logarithmic scales at low rates of shear extending downward to  $1 \text{ sec}^{-1}$  the result was a straight-line plot with a slope of unity. This indicated Newtonian behavior, and the corresponding values of reduced viscosity, when extrapolated to zero concentration, yielded an intrinsic viscosity of  $27 \text{ dl/g}$ . To corroborate these results, further viscosity measurements were carried out on solubilized collagen preparations using a ZIMM-CROTHERS<sup>2</sup> viscometer, as manufactured by Beckman\*, over the range of rate of shear from  $0.02$ – $0.1 \text{ sec}^{-1}$ .

Two collagen preparations were used in this study. These were made by solubilizing diced calfskin corium in citrate buffer at pH 3.44, and  $I$ , 0.64. Preparation and verification followed methods described in the first paper. Assay of these preparations differed from the original procedure in that after dialyzing a definite volume of each collagen preparation to exhaustion of the electrolyte, twice that volume of acetone was added to the slurry before separation of the precipitated collagen. This caused precipitation of material that formerly remained in the supernatant liquid, and thus indicated somewhat higher values of collagen concentration.

The shearing stress for each observation with the ZIMM-CROTHERS viscometer was set by adjusting the altitude of the viscometer chamber relative to the rotating magnet. At each elevation the shearing stress was determined by using a solution of glycerine–water of known absolute viscosity. The average rate of shear is given by  $OKA^3$ :

$$G = \frac{1}{b-a} \frac{2\omega}{1/a + 1/b} \quad (1)$$

where  $G$  = rate of shear,  $b$  = radius of viscometer stator,  $a$  = radius of viscometer rotor, and  $\omega$  = angular velocity of viscometer rotor.

Since the glycerine–water solution is a Newtonian liquid, shearing rates can be calculated directly from

$$\tau = G\eta \quad (2)$$

where  $\tau$  = shearing stress and  $\eta$  = absolute viscosity of the glycerine–water solution.

In these experiments the viscometer stator and rotor radii were 6.62 and 4.90 mm, respectively, and the glycerine–water solution had an  $\eta = 8.12 \text{ cP}$ . All runs were at a temperature of  $25^\circ$ .

The value of shearing stress as calculated from Eqns. 1 and 2 is dependent only on the distance between the viscometer rotor and the rotating magnet<sup>4</sup> provided the drive motor is rotating at a speed greater than 500 rev./min. Therefore, after the shearing stress for a given altitude of the viscometer was determined, the instrument

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was held in this position and the standard glycerine-water solution was replaced with a collagen solution whose volume was carefully adjusted so that the viscometer rotor floated at the same level as before as gauged by cathetometer. A new value of  $\omega$  of the rotor was observed and applied to Eqn. 1 to give the average rate of shear for the collagen sample.

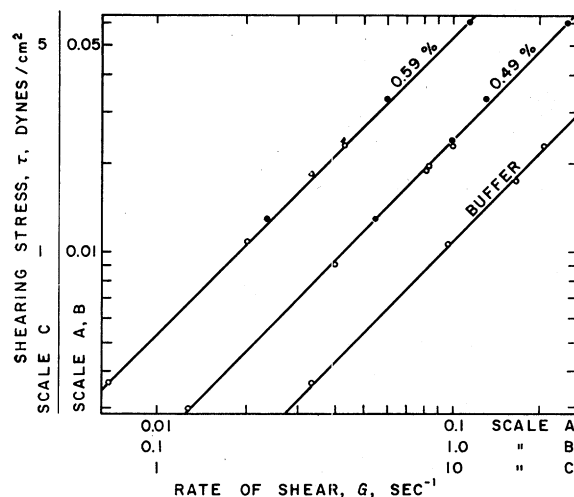


Fig. 1. Plot of shearing stress with respect to rate of shear for calfskin collagen solubilized in citrate buffer at pH 3.44. O, data obtained with ZIMM viscometer, scale A; also data for buffer, scale B; ●, data obtained with capillary viscometers, scale C.

The data are plotted in Fig. 1. The concentration of the two solubilized collagen preparations used are given above the respective plotted lines, and data for the buffer used as solubilizing agent are also given. Values of shearing stress and rate of shear at the capillary wall were obtained for the same collagen solutions in capillary viscometers and are included to show continuity between the two techniques. In drawing the graph, several logarithmic decades have been telescoped to one for the sake of compactness.

Each plotted line is straight and has a slope of unity (within 1%) and thus shows Newtonian behavior and confirms the conclusions drawn from the previous experiments carried out in capillary viscometers<sup>1</sup>.

Eastern Utilization Research and Development Division,  
Agricultural Research Service, U.S. Department of Agriculture,  
Philadelphia, Pa. (U.S.A.)

LEO D. KAHN  
LEE P. WITNAUER

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